

vasodilatation ($P<0.03$) and resulted in higher plasma t-PA antigen and activity concentrations during bradykinin infusion (11.3 ± 0.8 vs 6.8 ± 0.5 ng/mL and 16.5 ± 3.9 vs 6.6 ± 2.0 IU/mL at peak bradykinin dose; $P<0.002$) and a doubling of estimated net t-PA release ($P<0.05$). **Conclusion:** Intra-arterial TNF- α causes an acute local vascular inflammation associated with a substantial and sustained increase in local t-PA and IL-6 release. TNF- α also impairs endothelium-dependent vasomotion and augments acute endothelial t-PA release. These findings indicate that TNF- α has potentially adverse and beneficial effects on endothelial and vascular function.

2:45 p.m.

856-4

iNOS is a Mediator of Increased Arterial Intimal Thickening Induced by Passive Cigarette Smoke Exposure in Mice

Takeo Anazawa, Paul C. Dimayuga, Hongyan Li, Previn de Silva, Juliana Yano, Kuang-Yuh Chyu, Prediman K. Shah, Bojan Cercek, Cedars-Sinai Medical Center, Los Angeles, CA

Background: Active and passive smoking was associated with increased intimal/medial thickening in the Atherosclerosis Risk In Communities study, but molecular mechanisms contributing to this risk are incompletely understood. We evaluated the effect of passive smoke on arterial response to injury, and the potential role of iNOS gene in smoking induced effects on the arterial wall using iNOS $-/-$ mice.

Methods: Vascular injury was induced by placing a cuff around the right carotid artery. Wild type mice and iNOS $-/-$ mice of the same background were exposed to passive smoke (1 cigarette/day) or filtered room air. Expression of iNOS and PCNA in the arterial wall 3 days after injury was determined by immunostaining. Nitrate and nitrite (NOx) level 3 days after injury was measured by Griess reaction. Intimal thickening was measured 21 days after injury.

Results: iNOS expression in wild type mice exposed to passive smoke increased compared to mice exposed to room air, and was not detected in iNOS $-/-$ mice. Intimal thickening in iNOS $-/-$ mice exposed to passive smoke was profoundly reduced compared to wild type mice exposed to passive smoke (Table).

Medial areas were similar in all groups of mice.

Conclusion: Our results suggest that iNOS expression is a key mediator in the augmented response to injury in mice exposed to cigarette smoke. iNOS may mediate vaso-occlusive effects of exposure to cigarette smoke.

Table:

Groups	Wild-type + Room air	Wild-type + Passive smoke	iNOS $-/-$ mice + Room air	iNOS $-/-$ mice + Passive smoke
PCNA positive nuclei	8.1 ± 2.7 (n=5)	$15.7 \pm 5.2^*$ (n=5)	ND	4.0 ± 4.0 (n=3)
NOx level (μ M)	22.3 ± 7.3 (n=6)	$39.1 \pm 12.8^*$ (n=5)	$14.3 \pm 6.8^*$ (n=6)	12.5 ± 8.6 (n=6)
Intimal Area ($\text{mm}^2 \times 10^{-3}$)	9.2 ± 7.5 (n=6)	$23.3 \pm 12.9^*$ (n=5)	$5.5 \pm 4.7^*$ (n=5)	2.1 ± 1.6 (n=5)

* $p<0.05$ vs iNOS $-/-$ +Passive smoke, * $p<0.05$ vs Wild-type+Room air, * $p<0.05$ vs Wild-type+Passive smoke; ANOVA

3:00 p.m.

856-5

Cardiovascular Effects of the Endogenous Nitric Oxide Synthase Inhibitor Asymmetric Dimethylarginine (ADMA) and Evidence for ADMA Metabolism in Humans In Vivo

Vinod Achan, Michael Broadhead, Mohammed Malaki, James Leiper, Raymond MacAllister, Patrick Vallance, University College London, London, United Kingdom

BACKGROUND: Plasma levels of an endogenous NOS inhibitor, asymmetric dimethylarginine (ADMA), are elevated in chronic renal failure, hypertension, and atherosclerosis. ADMA levels are also significantly raised in patients with chronic heart failure (CHF) and in animals with CHF induced by coronary artery ligation or rapid pacing. Despite these observations the cardiovascular effects of a systemic increase in ADMA have not been studied in humans. **METHODS:** In a randomised, double-blind, placebo-controlled study using healthy male volunteers, we compared the effects of intravenous low dose ADMA and placebo on heart rate, blood pressure, cardiac output and systemic vascular resistance (SVR) at rest and during exercise. We also tested the hypothesis that ADMA is metabolised extensively in humans in vivo by a family of dimethylarginine dimethylaminohydrolase (DDAH) enzymes. **RESULTS:** Low dose ADMA reduced heart rate by a maximum of $9.2\pm 1.4\%$ from 58.9 ± 2.0 beats per min ($P<0.001$), and reduced cardiac output by a maximum of $14.8\pm 1.2\%$ from 4.4 ± 0.3 L/min ($P<0.001$). ADMA also increased mean blood pressure by a maximum of $6.0\pm 1.2\%$ from 88.6 ± 3.4 mm Hg ($P<0.005$), and increased SVR by a maximum of $23.7\pm 2.1\%$ from 1639.0 ± 91.6 dynes.s.cm $^{-5}$ ($P<0.001$). Handgrip exercise increased cardiac output in control subjects by a maximum of $96.8\pm 23.3\%$. In contrast, those subjects given ADMA increased their cardiac output by $35.3\pm 10.6\%$ during exercise, representing a significant reduction in the cardiac response to exercise ($P<0.05$). DDAHs metabolise ADMA to citrulline and dimethylamine (DMA). Urinary DMA:Cr ratios significantly increased from 1.26 ± 0.32 to 2.73 ± 0.59 following ADMA injection ($P<0.01$). We estimate from this that humans generate approximately 300 μ mol of ADMA per day, of which approximately 250 μ mol is metabolised by DDAHs. **CONCLUSIONS:** This study defines the cardiovascular effects of a systemic increase in ADMA in humans. These are similar to changes seen in diseases associated with ADMA

accumulation, suggesting that ADMA could directly contribute to their pathogenesis. Finally, our data also indicate that ADMA is metabolised by DDAHs extensively in humans in vivo.

3:15 p.m.

856-6

Levels of Nitrotyrosine, an Inflammatory Marker Generated by Nitric Oxide Derived Oxidants, Is Associated With Risk of Coronary Artery Disease

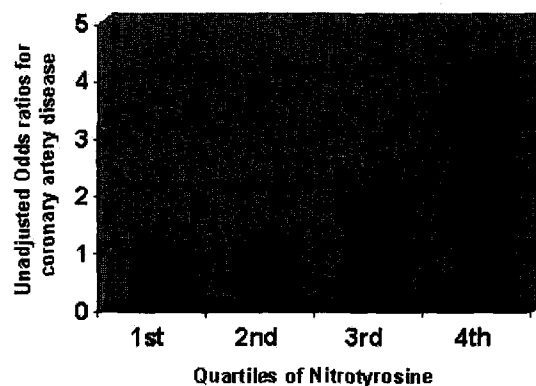
Mehdi H. Shishehbor, Ronnier J. Aviles, Marie-Luise Brennan, Xiaoming Fu, Marc S. Penn, Dennis L. Sprecher, Noyan Gokce, John F. Keaney, Jr., Joseph A. Vita, Stanley L. Hazen, The Cleveland Clinic Foundation, Cleveland, OH. Boston University School of Medicine, Boston, MA

Background: Formation of nitric oxide (NO)-derived oxidants may serve as a mechanism linking inflammation to development of atherosclerosis. Nitrotyrosine, a specific marker for protein modification by NO-derived oxidants, is enriched in human atherosclerotic lesions and LDL recovered from human atheroma. Whether systemic levels of nitrotyrosine predict coronary artery disease (CAD) is not known.

Methods: Serum nitrotyrosine levels in 262 consecutive patients at a major metropolitan medical center were determined by mass spectrometry and correlated with the prevalence of CAD.

Results: The median nitrotyrosine content of plasma proteins was significantly higher in the CAD group (9.13 μ mol/mol vs. 5.66 μ mol/mol, $P<0.001$). Subjects in the upper quartile of nitrotyrosine levels had higher risk of CAD (unadjusted odds ratio, 4.06 ; 95% confidence interval, 1.94 to 8.50 ; $P<0.001$). After adjusting for Framingham risk factors and high sensitive C-reactive protein, upper quartiles of nitrotyrosine remained predictive for CAD risk (odds ratio, 3.01 ; 95% confidence interval, 1.28 to 7.07 ; $P<0.001$).

Conclusion: Elevated levels of nitrotyrosine, a specific protein modification produced by NO-derived oxidants and which is linked to CAD pathogenesis, serves as a significant and independent predictor of CAD risk. These results support a potential role for NO-derived oxidants as an inflammatory mediator in CAD and may have important implications for atherosclerosis diagnosis and risk assessment.



ORAL CONTRIBUTIONS

857 Vascular Diseases: Clinical Insights and Clinical Trials

Tuesday, April 01, 2003, 2:00 p.m.-3:30 p.m.
McCormick Place, Room S405

2:00 p.m.

857-1

Incidence and Characteristics of Ruptured Plaque in Femoro-Popliteal Arteries

Yoichiro Hongo Ali Hassan, Krishnankutty Sudhir, Daniel Adelman, Yasuhiro Honda, Paul G. Yock, Peter J. Fitzgerald, Stanford University, Stanford, CA, Pharmacocyclics Inc., Sunnyvale, CA

Background: Numerous studies have reported the characteristics of atherosclerotic lesions with plaque rupture in coronary arteries. However the incidence and characteristics of plaque rupture in the peripheral circulation have not been well studied.

Methods: Ninety-seven lesions in 40 patients scheduled for elective angioplasty in either the femoral or popliteal arteries were enrolled. IVUS was performed before intervention. Lesion inclusion criteria were: (1) segmental, (2) proximal reference % plaque area $< 50\%$, (3) degree of calcified arc $< 90^\circ$. Discontinuity of luminal surface with a cavity within the plaque mass was defined as plaque rupture. Lumen area, vessel area, degree of calcified arc, plaque eccentricity index (EI) and remodeling index (RI: lesion / proximal reference vessel area) were measured.

Results: Plaque rupture was observed in 42 lesions (43%). When lesions with and without plaque rupture were compared, lumen area, degree of calcified arc and EI were identical (8.9 ± 3.7 vs 8.9 ± 5.2 mm 2 , 22 ± 25 vs $25\pm 31^\circ$, 0.74 ± 0.18 vs 0.73 ± 0.18 , NS). However, vessel area and RI were significantly higher in lesion with plaque rupture (30.1 ± 6.0 vs 26.0 ± 6.9 mm 2 , $p=0.001$, 1.09 ± 0.19 vs 0.98 ± 0.18 , $p=0.008$). The distribution of remod-